

INFORMATION REPORT INFORMATION REPORT

CENTRAL INTELLIGENCE AGENCY

50X1-HUM

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COUNTRY Poland

REPORT

SUBJECT Research Project - Environmental Stress as a Contributing Factor in Animal Diseases

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THIS IS UNEVALUATED INFORMATION

the 1963 annual report of [10 pages, English] a Polish research project conducted at the Institute for Veterinary Research at Pulawy. The project, which is under the direction of Dr Teodor Tuskiewicz is entitled "Environmental Stress as a Contributing Factor in Animal Diseases." The summary of progress reported is as follows:

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1. We have found in our previous works that after application of ACTH/Depot-Acethropan, Hoechst/ in doses of 40 I U, the 12-weeks-old cockerels showed a distinct functional excitation of adrenal cortex. As the action of ACTH in poultry is a subject of many controversial reports, we made a series of our own investigations. The cockerels were given ACTH intramuscularly in large /40 I U/ and small /4 I U/ doses, as well as the preparation having been introduced into the heart blood. A number of biochemical parameters was examined during the 36 hours from the time of application of the ACTH to cockerels. No great differences in the action of ACTH have been found after its application in various doses or various ways. It seems, that some further studies are necessary to explain the action of ACTH in hens.

2. The effects of viral /Newcastle Disease Virus/ and bacterial /Pasteurella multocida/ influence upon the behaviour of many biochemical parameters in chickens, were examined on a large quantity of cockerels. It has been found that the viral infection, as well as the bacterial infection, produces in organism several biochemical changes, which suggest the functional stimulation of adrenal glands. Due to the action of Prednisolone and also owing to the influence of such factors as high temperature of environment or shaking of chickens on the laboratory shaking machine, this process of infection was accelerated and the death rate of chickens went up.

3. In the reported year, 393 chicks were used in the experiments and the total number of biochemical determinations was approximately 5800.

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ANNUAL REPORT
OF THE RESEARCH CONDUCTED UNDER GRANTS AUTHORIZED BY PUBLIC LAW 480.

1. NAME AND ADDRESS OF THE RESEARCH INSTITUTION: The Institute
for Veterinary Research, Puławy, Poland.
2. PRINCIPAL INVESTIGATOR IN CHARGE: Docent Dr. Teodor JUSZKIEWICZ.
3. PROJECT TITLE: Environmental Stress as a Contributory Factor
in Animal Diseases.
4. PROJECT NUMBER: E 21-ADP-7
5. GRANT NUMBER: FG-Po-134-62.
6. REPORT PERIOD: From January 1st, 1963 to December 31st, 1963.

Signature
of the Principal Investigator
in Charge

T. Juszkiwicz
Docent Dr. Teodor JUSZKIEWICZ

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of the Research Institution

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Professor Dr. Stanisław KRAUSS

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SUMMARY OF PROGRESS.

We have found in our previous works that after application of ACTH/Depot-Acethropan, Hoechst/ in doses of 40 I.U., the 12-weeks-old cockerels showed a distinct functional excitation of adrenal cortex. As the action of ACTH in poultry is a subject of many controversial reports, we made a series of our own investigations. The cockerels were given ACTH intramuscularly in large /40 I.U./ and small /4 I.U./ doses, as well as the preparation having been introduced into the heart blood. A number of biochemical parameters was examined during the 36 hours from the time of application of the ACTH to cockerels. No great differences in the action of ACTH have been found after its application in various doses or various ways. It seems, that some further studies are necessary to explain the action of ACTH in hens.

The effects of viral /Newcastle Disease Virus/ and bacterial /Pasteurella multocida/ influence upon the behaviour of many biochemical parameters in chickens, were examined on a large quantity of cockerels. It has been found thatt the viral infection, as well as the bacterial infection, produces in organism several biochemical changes, which suggest the functional stimulation of adrenal glands. Due to the action of Prednisolone and also owing to the influence of such factors as high temperature of environment or shaking of chickens on the laboratory shaking machine, this process of infection was accelerated and the death rate of chickens went up.

In the reported year, 393 chicks were used in the experiments and the total number of biochemical determinations was approximately 5800.

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DETAILED REPORT

I. EFFECTS OF ACTH-ACTION IN CHICKENS.

It has already been mentioned in the previous report that the action of ACTH in chickens was a subject of controversy in the available scientific literature. A short review of literature in this respect was included into our "Annual Report 1962" and presented in the paper "Pathophysiology of the Pituitary-Adrenal Axis in Birds" /3, 4, 5/.

In our previous investigations we have found that after intramuscular application of ACTH /Depot, Acethropan, Hoechst/ in doses of 40 I.U., the 12-weeks-old cockerels used to show, among others, a reaction similar to that which could be observed in such cases in mammals /3, 4, 5/. In some later experiments, however, done with the preparations of other firms, we have not been able to find the confirmation of this observation. Therefore, it was of interest to re-investigate the action of ACTH in chickens within a specially designed experiment.

Material and Methods

E x p e r i m e n t I. Seventy two 11-weeks-old cockerels had been subdivided into three groups. Those of group No.1 were given single intramuscular injection of 40 I.U. ACTH /Polfa/ per bird; group No.2 - 4 I.U. ACTH per cockerel, and the group No.3 /control/ - saline solution.

After 1, 6, 12 and 24 hours respectively from the time of injection of the preparation, 6 cockerels from every group were killed and the material for analyses was collected. The following determinations have been made: weighing of adrenal glands, spleen and thymus; concentration of adrenal and blood ascorbic acid; concentration of adrenal and blood cholesterol; concentration of liver glycogen and blood glucose; and concentration in blood serum of sodium, potassium, calcium, inorganic phosphorus and magnesium. The methods of determinations were given in the "Annual Report 1962".

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Experiment II. One hundred and ten 12-week-old cockerels were divided into three groups. The cockerels of group No.1 were given 4 I.U. of ACTH per bird, injected into the heart blood; Those of group No.2 - 4 I.U. of ACTH per bird, intramuscularly; and those of group No.3 /control/ - 4 ml of sterilized water, the same as used for dissolving the ACTH, into the heart blood.

Then after 1, 6, 12, 24 and 36 hours from the time of application the preparations, an appropriate number of birds from every group was killed and the material collected for analyses. All the same determinations as in the Experiment I were made, with an addition that also the concentration of liver cholesterol was determined.

Results and Discussion

It was found in the Experiment I, that the applied preparation of ACTH /Polfa/ induced rather faint changes in the examined parameters. Most of the found changes were more distinct at the beginning of the experiment. Those chickens which were given 4 I.U. of ACTH, showed after 1 hour, a decrease in weight of the spleen and thymus and also a decrease in contents of adrenal cholesterol. Instead, and what was rather interesting, some increase in ascorbic acid was noted in the adrenals and a decrease in weight of the adrenals. However, after the tenfold doses of ACTH, i.e. after 40 I.U., certain decrease of ascorbic acid concentration in the adrenals was noted together with the increase in weight of adrenals. These changes were statistically significant only after 1 hour of ACTH action. The other examined parameters did not present any significant differences.

It is, however, to be emphasized that in spite of such differing doses being compared as 4 I.U. and 40 I.U. per specimen, the differences in response were relatively minor. When analyzing these results we were rather surprised with the limited activity of the applied preparations. Especially, that when in our previous experiments the long-acting ACTH /Depot-Acethropan, Hoechst/ was used, a significant reaction to 40 I.U. of ACTH was found. It seemed therefore, that these results would confirm the assumption of Garren et al./2/, Zarrow et al./1/ and of some other investigators, that the avian tissues cause a rapid destruction of the ACTH molecule.

With that point of view in mind it has been decided to administer the ACTH in the next experiment intravenously in doses of 4 I.U. For that purpose the technique has been adopted to introduce the drug

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directly into the heart. At the same, in another group of cockerels, the same preparation of ACTH was administered intramuscularly, and the control group was given only the sterilized water into the heart blood.

The obtained results show that in the chickens which were given injections of ACTH intramuscularly, the examined parameters behaved similarly to those in Experiment I. And to our surprise, the effects of ACTH, when administered directly into the heart blood, did not much differ from its action after the intramuscular application. Although our investigations have been conducted on a comparatively large number of cockerels, we think it is still too early to draw any definite conclusions. At present, we are considering the possibility to make a similar experiment on chickens with the ACTH made by Hoechst. We shall also endeavour to test the activity of extracts prepared from the anterior pituitary of chickens. For, it seems, that the ACTH action in poultry may chiefly depend upon the composition of ACTH and upon the initial structure of the amino acids found in corticotrophins as well. It is a known fact that the preparations of various firms differ widely in this respect. Perhaps this is the reason of such a discrepancy in the reported effects of ACTH action in poultry.

Our point of view is that no sooner than after the completion of just mentioned additional experiments, the up to now results of our investigations will be ready for closer analysis and their publication.

II. EFFECTS OF VIRAL INFECTION STRESS ON PITUITARY-ADRENAL AXIS IN CHICKENS.

Asdel and Hanson found that the increase in body virus titer of Newcastle disease virus was usually interrupted at about the 36th hour and this interruption lasted 12 to 24 hours. Injection of hydrocortisone usually eliminated the period of interruption and shortened the period between inoculation and appearance of clinical signs and eventual death /6/.

In our experiments we wanted to determine to what extent the changes caused by the viral infection in an organism are similar to those which are noted under the influence of corticosteroid hormones action. At the same time we want to inquire into the question of whether the large doses of Prednisolone would influence the development of viral infection in chickens.

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Material and Methods

The experiment was conducted on 145 eleven-week-old cockerels of Pol-bar breed. From that number 17 untreated cockerels were allocated at random for the starting point determinations, or normal values of the examined parameters. The remaining cockerels were divided into three groups.

Group No.1 amounted to 48 chickens. Every chicken of this group was inoculated intramuscularly with 0.2 ml of 10^{-14} dilution of the highly virulent field strain of Newcastle disease virus /NDV/. After 12, 24, 48 and 72 hours from the time of infection, 8 cockerels were selected at random, killed and the representative body tissues or fluids were taken for examinations. After the remaining 18 chickens had been kept for 100 hours, 50 per cent of them died from the virus infection and then 9 surviving chickens were killed for examination.

Group No.2 of 40 chickens was given intramuscularly 10 mg of Prednisolone acetate /Hostacortin H, Hoechst/ per cockerel twice, i.e. the first time 24 hours before the time of infection of the first group and then, the second time at the same time when the first group was being infected. Then, after 6, 12, 24 48 and 72 hours from the second injection of Prednisolone, 8 chickens were selected at random every time for the chemical and virological determinations.

Group No.3 of 42 cockerels was given in the same manner and in the same doses the Prednisolone, and one hour later, after the second application of them preparation, the chickens were infected with the same dose of NDV as the chickens of the group No.1 The times of killing the chickens and collecting the material were 12, 24 and 48 hours from the time of infection. The remaining chickens of this group were killed, when half of their number died, what for this group happened 74 hours after the infection.

The following parameters have been determined with this experiment: body weight; weight of spleen, adrenals and thymus; concentration of adrenal- and blood cholesterol; concentration of liver glycogen and of blood glucose; and the concentration in blood serum of sodium, potassium, calcium, inorganic phosphorus and magnesium. The methods of determinations were quoted in the "Annual Report 1962".

Apart from that, the virus content in blood, brain, spleen, lungs and muscles was determined by chicken embryo inoculation.

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FIG. 1.

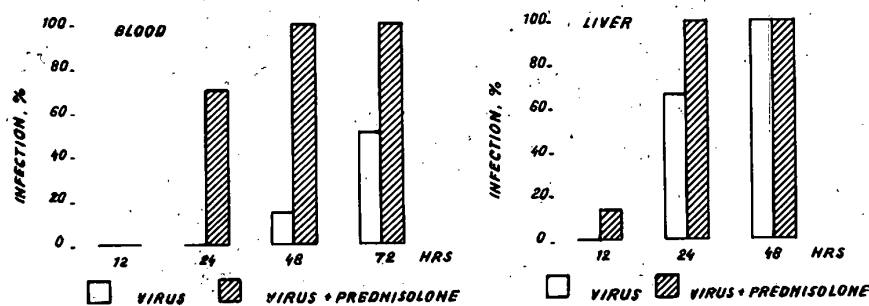
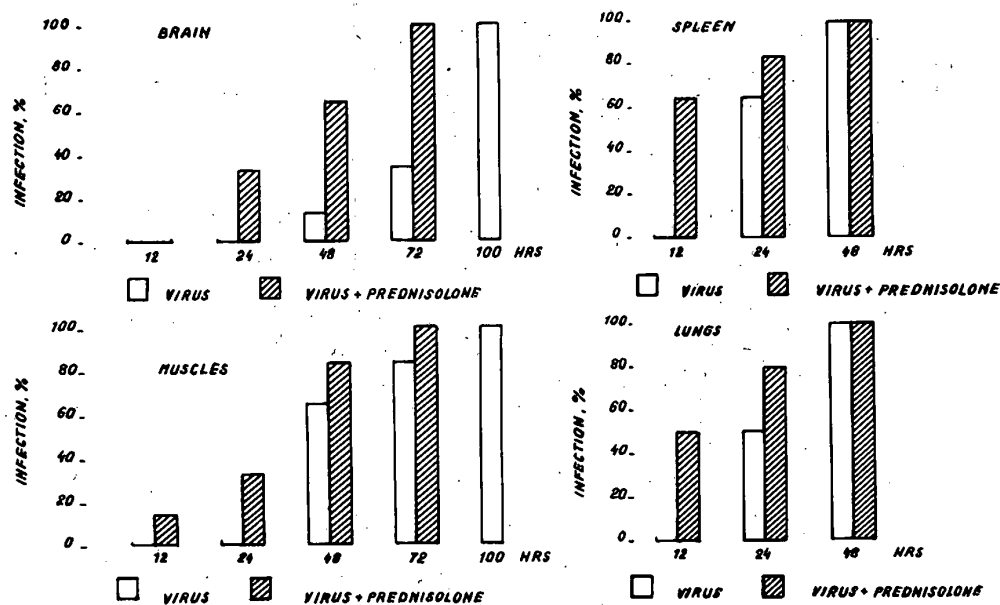


FIG. 2.



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Response of 11-week old cockerels to Newcastle diseases virus inoculation /VIR/, prednisolone injection /PR/, and a combined virus and prednisolone injection /VIR+PR/. All values are means \pm standard errors, P = compared with normal values /untreated subject/. Numbers in parenthesis represent number of animals studied.

Determinations	Normal values	Group	Time in hours				
			6	12	24	48	72
Spleen weight,	2.13	VIR /39/	-	1.81 \pm 0.20 P>0.2	2.48 \pm 0.20 P>0.2	3.33 \pm 0.29 P<0.005	2.34 \pm 0.25 P>0.5
g/kg body weight	± 0.13	PR /40/	0.90 \pm 0.06 P<0.001	0.86 \pm 0.08 P<0.001	0.86 \pm 0.09 P<0.001	0.66 \pm 0.05 P<0.001	0.75 \pm 0.07 P<0.001
		VIR+PR /33/	-	0.80 \pm 0.09 P<0.001	0.97 \pm 0.07 P<0.001	0.97 \pm 0.10 P<0.001	0.99 \pm 0.11 P<0.001
Thymus weight,	4.31	VIR /39/	-	2.18 \pm 0.34 P<0.001	3.64 \pm 0.39 P>0.1	3.30 \pm 0.34 P>0.1	2.88 \pm 0.41 P<0.05
g/kg body weight	± 0.29	PR /40/	2.03 \pm 0.19 P<0.001	1.78 \pm 0.12 P<0.001	1.61 \pm 0.29 P<0.001	0.70 \pm 0.10 P<0.001	0.96 \pm 0.16 P<0.001
		VIR+PR /33/	-	1.45 \pm 0.31 P<0.001	2.20 \pm 0.37 P<0.005	1.22 \pm 0.20 P<0.001	0.88 \pm 0.09 P<0.001
Adrenal weight,	99.1	VIR /39/	-	114 \pm 7.3 P>0.1	105 \pm 3.4 P>0.5	103 \pm 6.4 P>0.1	116 \pm 14.8 P>0.2
mg/kg body weight	± 4.64	PR /40/	90 \pm 5.3 P>0.2	86 \pm 6.1 P>0.1	101 \pm 7.2 P>0.5	95 \pm 6.5 P>0.5	91 \pm 4.4 P>0.2
		VIR+PR /33/	-	97 \pm 4.4 P>0.5	104 \pm 4.5 P>0.4	94 \pm 3.1 P>0.4	117 \pm 6.3 P<0.05
Ascorbic acid,	170.7	VIR /39/	-	184 \pm 53.6 P>0.4	160.7 \pm 15.5 P>0.2	149.8 \pm 10.8 P>0.05	147.0 \pm 3.8 P<0.005
adrenals,	± 5.89	PR /40/	225.6 \pm 8.2 P<0.001	197.8 \pm 6.3 P<0.05	178.0 \pm 6.9 P>0.5	165.6 \pm 8.9 P>0.2	119.1 \pm 5.8 P<0.001
mg/100 gm		VIR+PR /33/	-	203.1 \pm 5.5 P<0.01	158.6 \pm 11.8 P>0.2	139.2 \pm 16.4 P>0.05	171.6 \pm 23.8 P>0.5
Cholesterol,	55 \pm 25	VIR /39/	-	59.39 \pm 2.43 P>0.4	90.56 \pm 1.58 P<0.01	49.69 \pm 1.38 P<0.005	53.81 \pm 2.54 P>0.2
adrenals,	± 1.95	PR /40/	59.33 \pm 3.09 P>0.5	58.41 \pm 5.86 P>0.5	57.84 \pm 9.55 P>0.5	61.36 \pm 3.53 P>0.2	50.05 \pm 3.90 P>0.1
mg/gm		VIR+PR /33/	-	54.41 \pm 1.44 P>0.1	50.95 \pm 3.01 P>0.05	55.11 \pm 8.69 P>0.2	41.72 \pm 2.86 P<0.001
Glycogen,	13.05	VIR /39/	-	15.01 \pm 1.54 P>0.05	13.00 \pm 0.71 P>0.2	10.01 \pm 0.68 P<0.01	7.98 \pm 1.16 P<0.005
liver,	± 0.39	PR /39/	25.12 \pm 0.95 P<0.001	22.20 \pm 1.32 P<0.001	25.85 \pm 3.44 P<0.005	26.20 \pm 1.24 P<0.001	21.53 \pm 3.9 P<0.05
mg/gm		VIR+PR /33/	-	21.61 \pm 1.36 P>0.1	23.70 \pm 4.72 P<0.05	19.07 \pm 1.35 P<0.001	9.47 \pm 1.08 P<0.05
Ascorbic acid,	1.68	VIR /39/	-	1.50 \pm 0.07 P>0.5	1.82 \pm 0.06 P>0.5	1.86 \pm 0.11 P>0.1	1.63 \pm 0.15 P>0.5
blood,	± 0.06	PR /40/	2.03 \pm 0.11 P<0.025	1.50 \pm 0.08 P<0.05	1.72 \pm 0.08 P>0.5	1.52 \pm 0.05 P>0.5	1.58 \pm 0.05 P>0.5
mg %		VIR+PR /33/	-	1.35 \pm 0.08 P<0.005	1.74 \pm 0.08 P>0.5	1.47 \pm 0.10 P>0.05	1.24 \pm 0.09 P<0.001
Glucose, blood,	235.03	VIR /39/	-	218.7 \pm 9.4 P<0.05	221.7 \pm 7.8 P<0.025	181.0 \pm 34.6 P>0.05	207.6 \pm 9.5 P<0.001
mg %	± 3.68	PR /40/	591.5 \pm 34.2 P<0.001	698.3 \pm 12.9 P<0.001	700.6 \pm 12.7 P<0.001	696.0 \pm 18.1 P<0.001	311.3 \pm 12.2 P<0.001
		VIR+PR /32/	-	618.8 \pm 17.2 P<0.001	421.8 \pm 54.8 P<0.005	369.0 \pm 33.3 P<0.005	322.9 \pm 65.1 P>0.2
Cholesterol,	168.0	VIR /39/	-	194.2 \pm 11.5 P<0.05	157.2 \pm 10.4 P>0.5	125.4 \pm 13.3 P<0.025	110.4 \pm 12.9 P<0.001
serum,	± 5.17	PR /40/	300.0 \pm 13.3 P<0.001	376.4 \pm 17.2 P<0.001	330.2 \pm 13.8 P<0.001	447.7 \pm 30.1 P<0.001	405.0 \pm 12.3 P<0.001
mg %		VIR+PR /33/	-	353.0 \pm 10.8 P<0.001	335.7 \pm 25.7 P<0.001	244.5 \pm 20.3 P<0.005	162.6 \pm 10.3 P>0.5
Sodium,	350	VIR /31/	-	340.1 \pm 3.5 P>0.1	330 \pm 14.3 P>0.05	315 \pm 14.7 P<0.025	310 \pm 12.6 P<0.005
serum,	± 7.11	PR /39/	319 \pm 9.8 P<0.005	340 \pm 8.7 P>0.05	343 \pm 7.2 P>0.05	389 \pm 15.8 P>0.1	339 \pm 11.5 P>0.1
mg %		VIR+PR /33/	-	323 \pm 10.7 P<0.01	302 \pm 9.9 P<0.001	330 \pm 16.1 P>0.05	349 \pm 6.2 P>0.1
Potassium,	16.7	VIR /36/	-	21.0 \pm 0.8 P<0.001	18.5 \pm 1.5 P>0.05	13.9 \pm 0.5 P>0.05	14.3 \pm 1.7 P>0.5
serum,	± 0.85	PR /39/	20.6 \pm 0.2 P<0.005	21.1 \pm 0.9 P<0.001	19.2 \pm 1.6 P<0.05	18.8 \pm 0.7 P<0.005	16.1 \pm 1.3 P>0.05
mg %		VIR+PR /31/	-	21.8 \pm 1.1 P<0.001	17.9 \pm 0.4 P<0.005	17.0 \pm 0.6 P>0.05	21.2 \pm 1.1 P<0.001
Calcium,	11.4	VIR /31/	-	11.7 \pm 0.5 P>0.1	11.9 \pm 0.5 P>0.2	9.2 \pm 0.5 P<0.025	9.4 \pm 0.8 P<0.05
serum,	± 0.39	PR /39/	12.8 \pm 0.5 P<0.025	10.9 \pm 0.4 P>0.5	10.7 \pm 0.4 P>0.5	11.6 \pm 0.3 P>0.1	12.3 \pm 0.3 P<0.025
mg %		VIR+PR /31/	-	11.4 \pm 0.5 P>0.2	10.6 \pm 0.6 P<0.025	10.0 \pm 0.8 P>0.2	9.7 \pm 0.5 P>0.1
Magnesium,	2.46	VIR /39/	-	3.58 \pm 0.13 P<0.001	2.66 \pm 0.41 P>0.5	3.30 \pm 0.16 P<0.005	2.42 \pm 0.25 P>0.5
serum,	± 0.12	PR /40/	2.40 \pm 0.18 P>0.5	3.98 \pm 0.07 P<0.001	3.05 \pm 0.29 P<0.001	4.94 \pm 0.25 P>0.1	3.78 \pm 0.31 P<0.005
mg %		VIR+PR /33/	-	3.62 \pm 0.05 P<0.001	2.92 \pm 0.35 P>0.2	3.79 \pm 0.44 P<0.025	2.72 \pm 0.25 P>0.2
Phosphorous,	5.89	VIR /39/	-	6.42 \pm 0.2 P>0.05	6.6 \pm 0.2 P<0.025	5.8 \pm 0.3 P>0.5	7.32 \pm 0.6 P<0.05
serum,	± 0.11	PR /40/	4.8 \pm 0.3 P<0.01	4.2 \pm 0.3 P<0.001	4.4 \pm 0.1 P<0.005	4.3 \pm 0.2 P<0.001	4.8 \pm 0.3 P<0.01
mg %		VIR+PR /33/	-	4.1 \pm 0.1 P<0.05	4.7 \pm 0.3 P<0.01	4.3 \pm 0.2 P<0.001	6.34 \pm 0.1 P<0.001

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Results

The obtained results of biochemical parameters have been collected in the table. The invasion of selected tissues by the virus is shown in the figures 1 and 2.

Discussion

Contrary to the doubtful action of ACTH in chickens, the effect of Prednisolone action was very distinct. There was a considerable thymico-lymphatic involution, and a highly significant increase in blood glucose and liver glycogen /glyconogenesis/. The level of cholesterol, potassium and magnesium in the blood serum increased while the level of inorganic phosphorus decreased. These results are, taking generally, in agreement with those of the other investigators /1, 7, 8/. It should be pointed out that some time later, when the concentration of the introduced Prednisolone became more dilute, it was also possible to notice some, although a weak one, response from the side of the adrenals, viz: decrease in concentration of adrenal ascorbic acid and adrenal cholesterol.

With the viral infection, however, the examined parameters behaved differently. At the initial period, which was about 24 - 48 hours long, there was an increase in weight of spleen and thymus, but the number of other parameters did not differ from the normal values. Whereas, in the later period, the typical effects of the stress reaction were evident. There was a decrease in weight of the spleen and thymus, while the weight of the adrenals went up. There was a decrease in concentration of ascorbic acid and cholesterol in adrenals. However, it is necessary to point out the low reactivity of avian adrenals. The levels of cholesterol and glucose in blood went down, and also the concentration of glycogen in liver significantly decreased. One might here argue whether in this case we had to deal with the diminished production of glucocorticoids, or what seems to be of greater likelihood, with the more intense expenditure of energetic material by the organism at the time of viral infection.

Under the action of the two factors, i.e. of the NDV infection and of the Prednisolone, the examined parameters used to develop more or less intermediate values.

As it can be seen from the Fig. 1 and Fig.2 the invasion of NDV was strongly accelerated by Prednisolone in all the examined tissues. Fifty per cent of cockerels of the group No.3, which were inoculated

FIG. 3

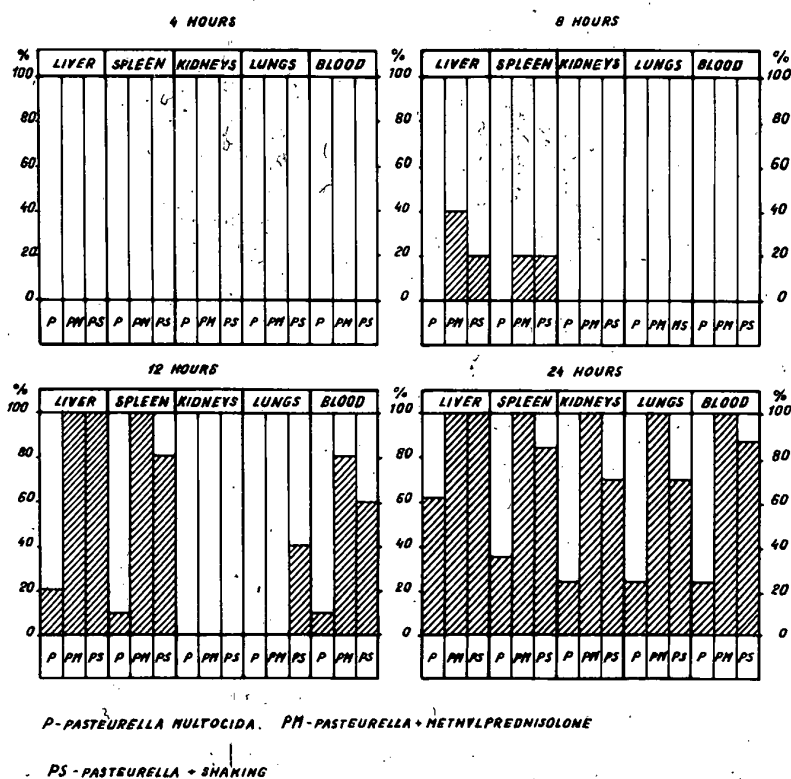
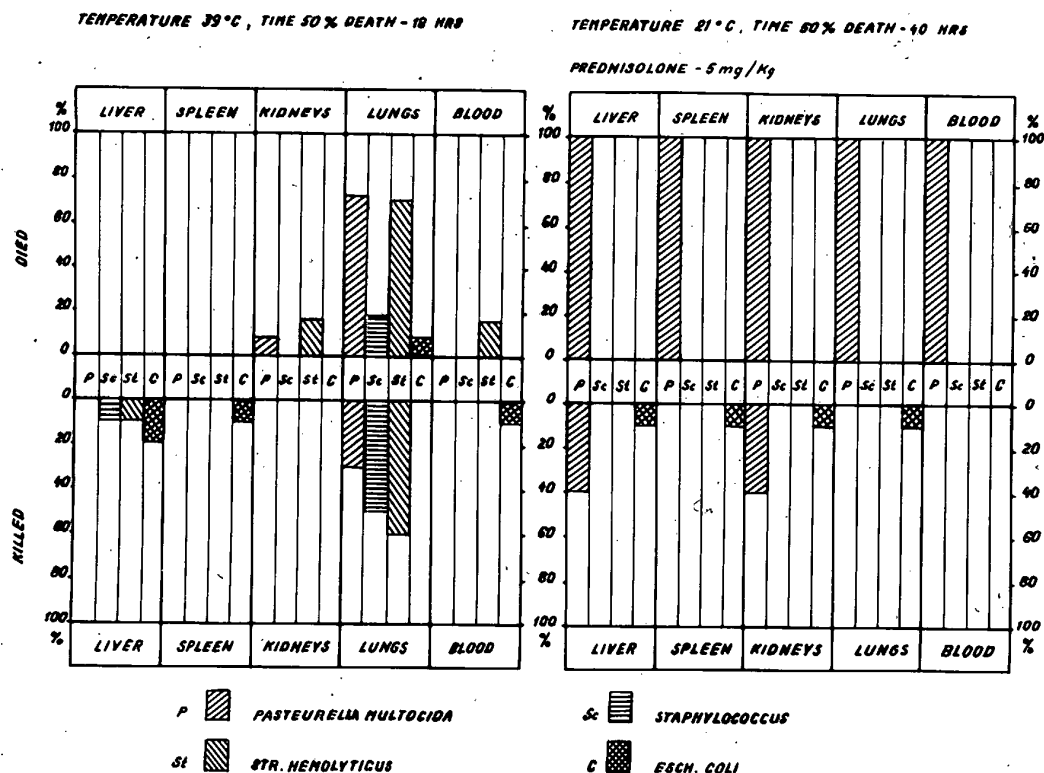


FIG. 4



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with the NDV and prior to that received intramuscularly the Prednisolone, died within 74 hours. During the same infection time, among the chickens of the group No.1, inoculated only with the virus /NDV/, hardly 6 per cent died. Fifty per cent of the cockerels of the group No.1 died within 100 hours of infection.

To complete this picture, it is certainly of some use to remind here that the similar course of action was also noticed in our experiments made earlier with the bacterial infection /ref.3 and the previous reports/.

The Fig.3 shows the results of bacteriological investigations in an experiment made on three groups of 12-week-old cockerels^{+/}. One group /P/ was infected with the virulent strain of *Pasteurella multocida*, the second group /PM/ - infected similarly but the chickens were given injections of 5 mg of Methylprednisolone twice, i.e. 24 hours and 1 hour prior to the onset of infection, the third group /PS/ - infected as the first one, but before the infection, the chickens spent 6 hours on the running laboratory shaking machine. After 4, 8, 12 and 24 hours from the time of infection the chickens were chosen at random from all groups, whereafter the material was taken for biochemical and bacteriological examinations. As it can be seen from the Fig.3, in chickens which were given the methylprednisolone, or were shaken, the *Pasteurella multocida* infection grew faster. Thus, perhaps, the fact is to be explained that after 24 hours from the infection, in the group /P/ which was infected with *Pasteurella multocida*, without any additional stressor factors, not a single cockerel died. Instead, in the group PM, infected with the same dose of bacteria, but premedicated with the methylprednisolone, 62 per cent of chickens died, and in the group /PS/, infected similarly but prior to that stressed with shaking, - 40 per cent of cockerels died.

In another experiment, investigations have been done,^{+/} which bring some additional information as to the relation between the *Pasteurella multocida* infection and the stress. Sixty six 11-week old cockerels of Polbará breed were infected with a slightly virulent strain of *Pasteurella multocida*, applying the dose which did not cause any clinical symptoms of infection. All the chickens were divided at random into three groups. The chickens of the first group were infected with *Pasteurella multocida* and an hour later placed in a heat chamber with the temperature 39°C and the relative humidity 60 per cent. In these conditions 50 per cent of chickens died after 18 hours. The chickens of the

^{+/} This paper will be published in 1964.

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second group were infected with the same dose of Pasteurella, but 25 hours and 1 hour prior to the infection, all of them got 5 mg/kg of Prednisolone acetate each. These chickens were kept in the room temperature of 20 - 21°C. After 40 hours 50 per cent of cockerels died.

The chickens of the third group were infected similarly like the others with the Pasteurella, but no other additional stress factors were applied to them. The chickens were kept at the room temperature of 20 - 21°C.

When 50 per cent of chickens died in the first two groups, the remaining, which survived, were killed and the material collected for biochemical and bacteriological determinations. In the third group, where no chickens died, 48 hours after the time of infection 10 chickens were selected at random and killed. The remaining 12 chickens were maintained for six months.

The fig.4 shows the results of bacteriological examinations in the two first groups of chickens which died and which were killed. The results of bacteriological investigations concerning the third group proved to be completely negative ones.

Conclusions

It looks as if on the ground of the results of our work, briefly outlined in this report, the following conclusions may be deduced:

1. The biological activity of adrenal cortex in poultry seems to be characterized by some partial independence from the anterior pituitary gland.
2. The mechanism of adrenocorticotrophin action in poultry is not clear up to now and it necessitates some further investigations.
3. Corticosteroid hormones, like all the other applied stress-factors i.e. temperature, shaking, bacterial infection, viral infection, cause in poultry a number of various biochemical changes. Their intensity, their course with the time and their character depend mainly upon the kind of stimulus.
4. The course of bacterial and viral infections may vary in a considerable manner due to the action of some additional stimuli from the environment.

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PLANS FOR THE FUTURE WORK.

1. Some additional experiments are planned to supplement the investigations on the action of ACTH in poultry, viz.:

- a/ to administer intravenously those preparations which proved to be effective with their intramuscular application;
- b/ to apply small doses of ACTH for longer periods of time - 14 days.

2. To conduct further investigations on the susceptibility of chickens to bacterial and viral infections when subjected to stressful conditions /heat, cold and transportation/.

3. Investigations on the action of compounds diminishing the stress effects in chickens and their therapeutical value against bacterial and viral infections in stressful conditions.

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